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Assessing Feeding Regimes and Its Impact on Tilapia (*Oreochromis* sp.) Performance and Aquaculturist Perceptions in Aquaculture

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ABSTRACT

This study evaluated the effect of four different feeding regimes on the growth, health, economic effiency and perception of aquaculturists regarding *Oreochromis* sp. in the Guayas Province, Ecuador. Four treatments based on different feeding regimes were established: T1, once a day with a daily interval; T2, once a day with an interval every two days; T3, twice a day with a daily interval; and T4, twice a day with an interval every two days. Growth parameters such as total fish weight gain, average daily weight gain, daily feed intake, expected weight gain for each daily feed intake, feed conversion ratio, protein efficiency, and protein productive value were measured. Health parameters included serum total protein content, albumin, serum globulins, cholesterol, triglycerides, and glucose levels. Economic efficiency parameters such as feed cost and feed cost per kilogram of weight gain were also assessed. Additionally, surveys were conducted with aquaculturists to assess the perception of aquaculture farmers regarding the importance of growth factors, fish health, and economic efficiency in the selection of feeding regimes for *Oreochromis sp*. The results indicated that treatment T3 was the most effective in maximizing growth; treatment T1 excelled in fish health; and treatment T2 was the most economically efficient. In conclusion, the choice of feeding regime will depend on the farmers' priorities, with a general trend, according to the survey, toward economic efficiency and growth, while fish health, although relevant, is a secondary priority.

INTRODUCTION

Tilapia (*Oreochromis* sp.) is one of the most important species in global aquaculture due to its rapid growth, resilience to diverse environmental conditions, and high commercial value **(Abd *et al.*, 2022)**. In Ecuador, particularly in the Guayas Province, tilapia production has gained significant economic importance, serving as a major source of income for local aquaculturists **(Jácome *et al.*, 2019)**. However, the success of tilapia farming largely depends on the selection of appropriate feeding regimes that maximize fish growth and health while optimizing operational costs **(Byamungu *et al.*, 2001; Kissinger *et al*., 2016)**.

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One of the main challenges faced by aquaculturists is determining the feeding regime that achieves an optimal balance between fish growth and economic efficiency. Feeding frequency and quantity are key factors that influence feed conversion rates, physiological health, and water quality **(Kojima *et al.*, 2015; Daudpota *et al*., 2016)**.
Despite the availability of various feeding strategies, it remains essential to evaluate how different regimes affect both the biological performance of fish and the aquaculturists' perception of their economic viability **(Riche *et al.*, 2004; Salas-Leiton *et al.*, 2008)**. The present study aimed to assess the effect of four different feeding regimes on the growth, health and economic efficiency of *Oreochromis* sp., as well as aquaculturists' perceptions regarding the importance of growth factors, fish health, and economic efficiency in the selection of feeding regimes. The regimes include feeding once a day with a daily interval (T1); once a day with an interval every two days (T2); twice a day with a daily interval (T3); and twice a day with an interval every two days (T4). Through an integrated approach combining experimental results and surveys of aquaculturists, this study intended to identify the potential of these feeding regimes for tilapia aquaculture in the region.

MATERIALS AND METHODS

Fish farming conditions

A total of 216 healthy tilapia (*Oreochromis* sp.) fry were used, sourced from the company Ecuahidrolizados, located in Guayas, Ecuador. The fish underwent a 20-day acclimatization process prior to the start of the experiment, during which they were fed a basal diet (Table 1). Additionally, health checks were conducted throughout this preparatory period according to the methology of **CCoA (2005)**. The fish were randomly distributed into 12 circular fiberglass tanks filled with dechlorinated tap water under constant aeration, with ambiental conditions being continuously monitored: temperature, pH, dissolved oxygen, ammonia-N, nitrite-N and water hardness. The water temperature was maintained at 26 ± 0.6°C and measured using a TP-03A calibrated digital thermometer, with daily readings to ensure accuracy. The water pH was controlled at 7.4 ± 0.2 using a PH100 portable digital pH meter, which was calibrated weekly to monitor water quality. Dissolved oxygen levels (7mg L^{-1}) were recorded using a DO-5509 portable dissolved oxygen meter, with a precision of ±0.1mg/ L. Weekly, ammonia-N levels (0.01 ± 0.02mg L^{-1}) and nitrite-N levels (0.02 ± 0.02mg L^{-1}) were measured using API aquaculture-specific colorimetric test kits. The water hardness (240-245mg L^{-1}) was determined using an API GH & KH automatic hardness test kit, ensuring that all parameters remained within the recommended ranges for tilapia farming. Additionally, the photoperiod in the laboratory was automatically controlled, providing 12 hours of light and 12 hours of darkness **(APHA, 1998)**.

Table 1. Proximate chemical composition of the basal diet

Ingredient ($g.kg^{-1}$)	
Yellow corn	208
Wheat flour	95
Wheat bran	55
Soy bean meal (40% CP)	260
Corn gluten (58 % CP)	100
Fish meal (60 % CP)	183
Fish oil	60
Methionine	4
Vitamins and minerals mixture [1]	3
Proximate chemical composition ($g.kg^{-1}$)	
Crude protein	337.2
Fat	93.8
Crude fiber	37.7
NFE [2]	409.5
GE ($MJ.kg^{-1}$) [3]	19.1
Lysine	17.3
Methionine	10.8
Ash	61.4
DM	939.7

[1]Composition of Vitamins and minerals mixture ($.kg^{-1}$): vitamin A (570000IU); vitamin B1 (54mg); vitamin C (0.15mg); vitamin E (680mg); vitamin B6 (32mg); biotin (47mg); calcium iodide (20mg); folic acid (81mg); manganese sulfate (60mg); sodium selenite (20mg); cobalt sulfate (560mg); iron sulfate (1500mg); copper sulfate (3100mg); calcium carbonate (till 1kg).
[2]Nitrogen-free extract (NFE): 100−(ash% + crude fiber%+ fat% +protein%)
[3]Gross energy(GE): Protein (22.5 $kJ.g^{-1}$), lipids (38.4 $kJ.g^{-1}$) and (16 $kJ.g^{-1}$)

Experimental design

Four feeding regime treatments were established with different feeding frequencies and intervals, as shown in Table (2). Three replicates were applied for each feeding regime (18 fish per replicate), with each replicate corresponding to one tank. Data collection was carried out over a period of 8 weeks. The diets were formulated according to standard measures for the species *Oreochromis* sp., compacted into 2.5mm pellets.

Table 2. Experimental design of the study

Treatment	Feeding frequency	Feeding intervals
Treatment 1 (T1)	Once a day	Daily interval
Treatment 2 (T2)	Once a day	Every two days
Treatment 3 (T3)	Twice a day	Daily interval
Treatment 4 (T4)	Twice a day	Every two days

Growth assessment

Fish weights were recorded at various stages of the process, initially at the start of the experiment, and then every two weeks. The growth performance of the fish was measured according to the methodology of **Castell and Tiews (1980)** using various parameters: Total weight gain, average daily weight gain, daily feed intake, expected weight gain for each daily feed intake, feed conversion ratio, protein efficiency ratio and protein productive value.

Total weight gain (TWG) was measured using Equation (1), where WT refers to the final weight of the fish, and WI refers to the initial weight of the fish.

$$\text{TWG (g/fish)} = \text{WT-WI}$$

Equation (1). Total weight gain

Average daily weight gain (ADWG) was measured using Equation (2), where TG refers to the total gain, and ED refers to the days of the experiment.

$$\text{ADWG (g/fish/day)} = \text{TG/ED}$$

Equation (2). Average daily weight gain.

Daily feed intake (IDA) was measured using Equation (3), where TFI refers to the total feed intake and NFD refers to the number of feeding days.

$$\text{IDA (g/fish/day)} = \text{TFI/NFD}$$

Equation (3). Daily feed intake.

Expected weight gain for each daily feed intake (EWG) was measured using Equation (4), where TWG refers to the total weight gain and NFD refers to the number of feeding days.

$$\text{EWG} = \text{TWG/ NFD}$$

Equation (4). Expected weight gain for each daily feed intake.

Feed conversion ratio (FCR) was measured using Equation (5), where *TFI* refers to the total feed intake, and *TG* refers to the total gain.

$$FCR\ (g/g) = TFI/TG$$

Equation (5). Feed conversion ratio

Protein efficiency ratio (PER) was measured using Equation (6), where TG refers to the total gain, and PI refers to the protein intake.

$$PER\ (g/g) = TG/PI$$

Equation (6). Protein efficiency ratio.

Protein productive value (VPP) was measured using Equation (7), where PG refers to the protein gain, and PI refers to the protein intake.

$$VPP = PG/PI$$

Equation (7). Protein productive value.

Fish health parameters

Blood samples were collected from a portion of the fish from each treatment at the end of the experiment, after 8 weeks. The serum, extracted without anticoagulant, was centrifuged for 20 minutes at 250 RPM. The obtained serum was used to determine biochemical blood indices such as total serum proteins, albumin, serum globulins, cholesterol, triglycerides, and glucose. Total serum protein (TSP) and albumin content (ALB) were determined by spectrophotometry using the Biuret method and the Bromocresol Green (BCG) method, respectively **(Reinhold *et al.,* 1953; Lumeij *et al.,* 1990)**. Serum globulins (SG) was estimated by subtracting albumin content from the total protein content **(Coles, 1986)**. Total cholesterol (CH), triglycerides (TRG), and glucose (GLU) levels were measured using colorimetric kits and spectrophotometry **(Trinder, 1969; Allain *et al.,* 1974; McGowan *et al.,* 1983)**.

Economic efficiency analysis

Economic efficiency was determined by measuring the following parameters according the methodology of **Dunning and Daniels (2001)**: Feed cost and Feed cost per kilogram of weight gain.

Feed cost (FC) was measured using Equation (8), where CD refers to the cost of one kilogram of each diet, and ATFI is the total feed intake (kg) during the experimental period (56 days).

$$FC = CD*ATFI$$

Equation (8). Feed cost.

The feed cost per kilogram gain (FCG) was measured using Equation (9), where TFC corresponds to the total feed cost and TWG is the total weight gain (kg)

$$FCG=TFC*TWG$$

Equation (9). Feed cost per kg gain

Data collection from the survey

In this study, a structured survey with closed-ended Likert scale questions (e.g., 'Very Important,' 'Important,' 'Neutral,' 'Slightly Important,' 'Not Important at All') was designed and administered to assess the perceptions of aquaculture farmers regarding the importance of growth factors, fish health, and economic efficiency in the selection of feeding regimes for *Oreochromis* sp. The survey measured perceptions of these three factors in relation to four specific feeding regimes: (1) T1, once daily with a daily interval, (2) T2, once daily with a two-day interval, (3) T3, twice daily with a daily interval, and (4) T4, twice daily with a two-day interval. A total of 150 aquaculture farmers, selected through convenience sampling, who applied one of these regimes on their farms in the province of Guayas, Ecuador, were surveyed. The surveys were distributed in person during farm visits, ensuring that a clear and detailed explanation of the study's purpose was provided. Additionally, informed consent was obtained from each participant, guaranteeing the confidentiality of their responses. Data collection took place over a two-month period, and frequency analysis was conducted to identify trends in farmers' preferences. Descriptive statistics were used to evaluate the perceived importance of each factor.

Statistical analysis

The obtained data were statistically analyzed using a one-way analysis of variance (ANOVA) to determine significant differences at $P< 0.05$, using R studio (version 4.4.2). Growth, fish health, and economic efficiency parameters were expressed as mean ± standard deviation (SD) from three replicates for each treatment. After identifying the appropriate statistical methods, Tukey's post-hoc test was applied with $\alpha = 0.05$. In the case of the survey, the quantitative results obtained were analyzed using descriptive statistics, such as frequencies and percentages, to identify trends in aquaculturists' perceptions of the different feeding regimes.

RESULTS AND DISCUSSION

Growth assessment

Table (3) shows the results on growth assessment where statistical analyses revealed significant differences among all treatments in the TWG parameter, where treatment T3 proved to be the most effective in maximizing weight gain in the fish, while treatment T2 was the least favorable. Regarding the ADWG parameter, feeding the fish once or twice daily (T1 and T3, respectively) was shown to be more efficient. In contrast, treatment T4 was the least effective for this parameter. As for the IDA parameter, treatment T2 showed the highest feed intake, followed by T4, although the latter had slightly lower intake. In the EWG parameter, treatments T2 and T4 recorded the highest values, with T4 being slightly superior, suggesting that feeding the fish every other day, regardless of frequency, optimizes weight gain, whereas daily feeding treatments were less efficient. In the FCR parameter, treatment T4 demonstrated the greatest efficiency, with the lowest value, while T3 had the highest value, indicating lower feed efficiency. Regarding the PER parameter, the highest protein efficiency was observed in treatment T1, comparable to that of T4, while treatment T3 showed the lowest protein efficiency. Finally, in the PPV parameter, treatment T3 was the most efficient in terms of protein utilization, with the highest value, whereas T4 had the lowest. Thus, treatment T3 was the most suitable for maximizing growth and the efficient use of resources in the fish compared to the other treatments.

Table 3. Growth assessment

T	TWG	ADWG	IDA	EWG	FCR	PER	VPP
T1	28.46 ± 0.38 ^{B}	0.38 ±0.01 ^{A}	0.55± 0.01 ^{D}	0.45± 0.004 ^{C}	1.53± 0.04 ^{D}	2.11± 0.03 ^{B}	1.11± 0.01 ^{D}
T2	18.57 ± 0.25 ^{D}	0.261± 0.02 ^{C}	0.91± 0.01 ^{A}	0.58± 0.002 ^{A}	1.73± 0.01 ^{A}	1.77± 0.004 ^{A}	0.95± 0.02 ^{A}
T3	29.95 ±0.41 ^{A}	0.399± 0.01 ^{A}	0.66± 0.01 ^{C}	0.43 ± 0.01 ^{C}	1.49± 0.02 ^{C}	1.91± 0.01 ^{C}	1.26± 0.03 ^{C}
T4	23.11 ± 0.31 ^{C}	0.321 ±0.01 ^{B}	0.87± 0.02 ^{B}	0.69± 0.01 ^{B}	1.43± 0.02 ^{B}	2.08± 0.01 ^{B}	1.18± 0.01 ^{B}

[1]T: Treatments. TWG: Total weight gain. ADWG: Average daily weight gain. IDA: Daily feed intake. EWG: Expected weight gain for each daily feed intake. FCR: Feed conversion ratio. PER: Protein efficiency ratio. VPP: Protein productive value.
[2]In each column, different letters (A, B, C, D) indicate significant differences between values ($P< 0.05$).

In terms of TWG and ADWG, the T3 treatment proved to be the most effective, which can be attributed to the ability of this regimen to maintain a constant availability of nutrients, promoting an optimal metabolic state that supports continuous growth **(Jobling, 1994; Yang *et al.*, 2019)**. Regarding the IDA parameter, the T2 treatment was the most efficient due to a compensatory hyperphagia effect, a phenomenon in which fish, following a period of food restriction, tend to consume more in subsequent meals to compensate for the lack of nutrients **(Wang *et al.*, 2000; Sayki *et al.*, 2020)**. The analysis of FCR showed that the T4 treatment was the most efficient, possibly because fish, with more time between meals, are able to digest and metabolize feed more completely, allowing for greater nutrient absorption, reducing waste, and optimizing the conversion of feed into body mass **(Ali *et al.*, 2003; Zheng *et al.*, 2015)**. The efficiency of the T1 treatment in the PER can be explained through previous research suggesting that moderate feeding frequency improves protein utilization efficiency **(Kaushik & Medale, 1994; Yengkokpam *et al.*, 2013)**. Finally, the high efficiency of the T3 treatment in terms of PPV suggests that fish fed more frequently are able to utilize proteins more effectively when energy is abundant, which is facilitated by the higher feeding frequency.

Fish health parameters

Table (4) shows the results on fish health parameters where in terms of TSP, treatment T1 proved to be the most effective, maintaining the highest levels of this parameter. In contrast, treatments T3 and T4 exhibited the lowest TSP levels, with no significant differences between them, making them less favorable compared to the other treatments. Regarding ALB levels, treatment T1 was again the most efficient in maximizing this parameter, while treatment T3 showed the lowest albumin levels. Concerning SG, treatment T1 also stood out, presenting the highest levels, whereas treatment T3 significantly reduced globulin levels. For the CH parameter, treatment T4 reached the highest levels, while treatment T2 presented the lowest levels. The remaining treatments showed intermediate levels with no significant differences. In the TRG parameter, significant differences were observed among all treatments, with treatment T4 being the most effective in maximizing triglyceride levels, while treatments T2 and T3 were the least effective. Finally, in the GLU parameter, treatment T2 exhibited the highest levels, whereas treatment T1 recorded the lowest. Overall, treatment T1 emerged as the most efficient for most health-related parameters in the fish, including TSP, ALB, and SG.

Table 4. Fish health parameters

T	TSP	ALB	SG	CH	TRG	GLU
T1	4.33± 0.03 ^{A}	2.88±0.01 ^{A}	1.43±0.01 ^{A}	105.49± 0.25 ^{B}	87.40±0.14 ^{B}	67.48±0.06 ^{D}
T2	3.83± 0.02 ^{B}	2.65±0.01 ^{B}	1.23±0.005 ^{B}	103.55± 0.05 ^{C}	85.70±0.1 ^{D}	76.47± 0.04 ^{A}
T3	3.51± 0.01 ^{C}	2.55±0.005 ^{C}	0.11±0.005 ^{D}	105.04±0.05 ^{C}	86.06±0.03 ^{C}	75.20± 0.03 ^{B}
T4	3.55± 0.01 ^{C}	2.58± 0.006 ^{D}	0.94± 0.05 ^{C}	114.58± 0.02 ^{A}	90.11±0.05 ^{A}	71.41± 0.03 ^{C}

[1]T: Treatments. TSP:Total serum protein. ALB: Albumin content. SG: Serum globulins. CH: Total cholesterol. TRG:Triglycerides. GLU: Glucose.
[2]In each column, different letters (A, B, C, D) indicate significant differences between values (P< 0.05).

In terms of TSP, T1 treatment was the most effective, maintaining the highest levels of this parameter. This can be explained by the fact that frequent and consistent feeding allows for optimal absorption and utilization of essential nutrients, which promotes protein synthesis in the body **(Lovell, 1989; Villarroel *et al.*, 2011)**. Regarding ALB levels, T1 treatment was again the most efficient. Albumin is a key protein in the transport of substances and osmotic regulation, and its synthesis is directly related to adequate nutrition and a balanced protein intake **(Kaushik & Luquet, 1980; Rios *et al.*, 2007)**. For SG, T1 treatment stood out, presenting the highest levels. Globulins are related to the immune system, and adequate, well-balanced feeding can enhance the immune response in fish **(Sheldon *et al*., 2002; Suvarna *et al.*, 2018)**. In terms of CH, T4 treatment reached the highest levels, indicating possible lipid mobilization from the diet or increased fat reserves due to longer intervals between meals **(Jobling *et al.*, 1994; Thongprajukaew *et al.*, 2017)**. Regarding TRG, T4 treatment stood out, suggesting that longer intervals between meals promote triglyceride accumulation as a form of energy storage **(Roberts, 1989; Ramos *et al.*, 2000)**. Finally, for GLU, T2 treatment exhibited the highest levels. This can be explained by the fish's response to an intermittent feeding pattern, which causes spikes in glucose levels as a response to food restriction followed by feeding **(Lehninger *et al.*, 2000; Shah *et al.*, 2017)**.

Economic efficiency analysis

Table (5) presents the results for economic efficiency parameters. The FC parameter indicates that treatment T2 was the most efficient, as it had the lowest value. In contrast, treatments T1 and T3 were the most expensive, with no significant differences between them. For the FCG parameter, treatment T2 also demonstrated the highest efficiency in maximizing yield relative to cost, while treatments T1 and T4 incurred the highest costs, reducing their efficiency. In conclusion, based on the results for both FC and FCG, treatment T2 stands out as the most economically efficient option. It not only has the lowest feed cost but also optimizes performance in relation to the cost per kilogram of weight gain.

Table 5. Economic efficiency analysis

T	FC	FCG
T1	0.42±0.005 ^{A}	1.21±0.006 ^{C}
T2	0.31±0.01 ^{C}	1.41±0.01 ^{A}
T3	0.43±0.005 ^{A}	1.34±0.006 ^{B}
T4	0.38±0.005 ^{B}	1.22±0.01 ^{C}

[1]T: Treatments. FC: Feed cost. FCG: Feed cost per kg gain.
[2]In each column, different letters (A, B, C, D) indicate significant differences between values (P< 0.05).

The analysis of the four treatments reveals strengths and weaknesses across various key parameters, allowing for the identification of the optimal treatment based on specific objectives. Treatment T1, feeding once a day with daily intervals, stands out in the parameters related to fish health, showing the highest levels of TSP, ALB, and SG, making it the most favorable option for maximizing health. However, this treatment is less efficient from an economic perspective, as it incurs high feed costs and lower efficiency in terms of EWG compared to the other treatments.

On the other hand, treatment T2, feeding once every two days, is the most efficient in terms of FC and FCG, making it the most cost-effective option. It also maximizes feed intake (IDA) and presents the highest GLU levels. Nevertheless, its main drawback is its low growth performance, being the least effective in terms of TWG and ADWG.

Treatment T3, feeding twice a day with daily intervals, is the best for maximizing growth, standing out as the most effective in TWG, ADWG, and PPV. However, this option is less economically efficient due to its high feed costs and negative impact on health parameters, showing lower levels of TSP, ALB, and SG. Additionally, it has the highest FCR, indicating lower feed efficiency.

Finally, treatment T4, feeding twice a day with intervals every two days, excels in FCR efficiency and also maximizes TRG and CH levels. However, it has significant disadvantages in ADWG, being the least favorable for this parameter, and it also shows lower levels of TSP, ALB, and SG, suggesting lower efficiency in maintaining fish health. Furthermore, this treatment incurs a high feed cost per kilogram of weight gain (FCG).

Analysis of survey results

Table (6) shows the descriptive values for the study population about the perceptions of aquaculture farmers regarding the importance of growth factors, fish health, and economic efficiency when selecting a feeding regime for *Oreochromis* sp., where responses are classified on a 1-to-5 scale (1= “Very important”, 2= “Important”, 3= “Neutral”, 4= “Slightly important”, 5= “Not important at all”), with “f” representing the frequency of each response.

The majority of respondents (70%) considered growth to be a 'very important' factor in their decision-making, followed by economic efficiency, with 73.3% of aquaculturists rating this aspect as 'very important.' On the other hand, fish health was perceived as 'very important' by 60% of aquaculturists, while 26.7% rated it as 'important.' Only a small percentage (2%) indicated that fish health was 'of little importance' or 'not important at all' in their decision-making. Regarding economic efficiency, this was the most highly valued factor, with 73.3% of respondents considering it 'very important,' while 20% rated it as 'important.' Only 2% viewed it as 'of little importance,' highlighting the tendency of aquaculturists to prioritize profitability in their operations. Finally, the results show that while all three factors are relevant, economic efficiency is the predominant factor in decision-making, closely followed by growth, reflecting the priorities of aquaculturists in maximizing both economic and production performance in their aquaculture systems.

Table 6. Perception of aquaculturists on key factors in selecting a feeding regimen for *Oreochromis* sp. (N=150)

Score	**1**	**2**	**3**	**4**	**5**	**Mean**	**Standard deviation**
Factor	f(%)	f(%)	f(%)	f(%)	f(%)		
Growth	105 (70)	35 (23.3)	5 (3.3)	3 (2)	2 (1.4)	4.58	0.78
Fish health	90 (60)	40 (26.7)	12 (8)	6 (4)	2 (1.3)	4.40	0.91
Economic efficiency	110 (73.3)	30 (20)	5 (3.3)	3 (2)	2 (1.4)	4.64	0.75

CONCLUSION

This study has demonstrated the potential of the four evaluated feeding regimens, identifying their strengths and weaknesses in relation to growth parameters, fish health, and economic efficiency. The choice of the most suitable feeding regimen for aquaculturists will depend on each producer's priorities. If the primary goal is to maximize growth, treatment T3 (feeding twice a day with daily intervals) is the most effective option. For those who prioritize fish health, treatment T1 (feeding once a day with daily intervals) is the most suitable. In terms of economic efficiency, treatment T2 (feeding once every two days) has proven to be the most cost-effective. Additionally, from the perspective of surveyed aquaculturists, economic efficiency ranks as the most valued factor when selecting a feeding regimen, followed by fish growth. While fish health is considered a relevant aspect, its priority is secondary compared to factors that directly impact profitability and productive performance. This finding reflects a tendency towards cost optimization and growth maximization, with fish welfare being important but not decisive in the decision-making process. The results of this study provide a comprehensive overview of how aquaculturists balance these three key factors, offering a solid foundation for adjusting feeding regimens according to the specific goals of each aquaculture operation.

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